

10/10/93  
FILING RECEIPT



UNITED STATES DEPARTMENT OF COMMERCE  
Patent and Trademark Office  
ASSISTANT SECRETARY AND COMMISSIONER  
OF PATENTS AND TRADEMARKS  
Washington, D.C. 20231

APPLICATION NUMBER	FILING DATE	GRP ART UNIT	FIL FEE REC'D	ATTORNEY DOCKET NO.	DRWGS	TOT CL	IND CL
08/073,577	06/07/93	1811	\$475.00	901104D	0	25	2

ALLEGRETTI & WITCOFF, LTD.  
10 SOUTH WACKER DR.  
CHICAGO, IL 60606

Receipt is acknowledged of this patent application. It will be considered in its order and you will be notified as to the results of the examination. Be sure to provide the U.S. APPLICATION NUMBER, FILING DATE, NAME OF APPLICANT, and TITLE OF INVENTION when inquiring about this application. Fees transmitted by check or draft are subject to collection. Please verify the accuracy of the data presented on this receipt. If an error is noted on this Filing Receipt, please write to the Application Processing Division's Customer Correction Branch within 10 days of receipt. Please provide a copy of the Filing Receipt with the changes noted thereon.

Applicant(s) RICHARD T. DEAN, BEDFORD, NH; BRIAN R. MOYER, BEDFORD, NH.

CONTINUING DATA AS CLAIMED BY APPLICANT-

THIS APPLN IS A CIP OF 08/019,864 02/19/93  
WHICH IS A CIP OF 07/653,012 02/08/91

\* SMALL ENTITY \*

TITLE

TECHNETIUM-99M LABELED PEPTIDES FOR IMAGING INFLAMMATION

PRELIMINARY CLASS: 514

**APPLICATION FOR UNITED STATES LETTERS PATENT  
IN THE UNITED STATES PATENT AND TRADEMARK OFFICE  
(Case No. 90,1104-D)**

**Inventor:** Richard T. Dean  
43 King Road  
Bedford, New Hampshire 03110

A Citizen of the United States of America

Brian R. Moyer  
3 Ascot Court  
Bedford, New Hampshire 03110

A Citizen of the United States of America

**Assignment:** Diatech, Inc.  
9 Delta Drive  
Londonderry, New Hampshire 03053

**Title:** TECHNETIUM-99m LABELED PEPTIDES FOR IMAGING  
INFLAMMATION

## TECHNETIUM-99m LABELED PEPTIDES FOR IMAGING INFLAMMATION

This is a continuation-in-part of U.S. Patent Application Serial No. 08/019,864, filed  
5 February 19, 1993, which is a continuation-in-part of U.S. Patent Application Serial No.  
07/653,012, filed on February 8, 1991.

### BACKGROUND OF THE INVENTION

10

#### 1. Field of the Invention

This invention relates to compositions that are radiolabeled scintigraphic imaging agents, methods of using these compositions and methods for producing such radiolabeled compositions. Specifically, the invention relates to technetium-99m labeled scintigraphic  
15 imaging agents that are compositions of a polysulfated glycan or mixture thereof and a compound comprising a polybasic moiety covalently linked to a radiolabel binding moiety radiolabeled with technetium-99m (Tc-99m). Methods and kits for making such compositions, and methods for using such compositions to image sites of infection and inflammation in a mammalian body are also provided.

20

#### 2. Description of the Prior Art

There is a clinical need to be able to determine the location and/or extent of sites of focal or localized infection and inflammation. In a substantial number of cases conventional methods of diagnosis (such as physical examination, x-ray, CT and ultrasonography) fail to  
25 identify such sites (e.g., an abscess). Although biopsy may be resorted to, it is preferable to avoid such invasive procedures, at least until they are diagnostically appropriate to identify the pathogen responsible for an abscess at a known location. Identifying the site of such "occult" infection is important because rapid localization and identification of the problem is critical to effective therapeutic intervention.

30 In the field of nuclear medicine, certain pathological conditions can be localized or the extent of such conditions determined by imaging the internal distribution of administered radioactively-labeled tracer compounds (i.e. radiotracers or radiopharmaceuticals) that

The preferred protecting group has the formula -CH<sub>2</sub>-NHCOR wherein R is a lower alkyl having 1 and 8 carbon atoms, phenyl or phenyl-substituted with lower alkyl, hydroxyl, lower alkoxy, carboxy, or lower alkoxy carbonyl.

Labeling with Tc-99m is an advantage of the present invention because the nuclear and radioactive properties of this isotope make it an ideal component of a scintigraphic imaging agent. This isotope has a single photon energy of 140 keV and a radioactive half-life of about 6 hours, and is readily available from a <sup>99</sup>Mo-<sup>99m</sup>Tc generator. Other radionuclides known in the prior art have effective half-lives which are much longer (*for example*, <sup>111</sup>In, which has a half-life of 67.4 h) or are toxic (*for example*, <sup>125</sup>I).

Each polybasic peptide-containing embodiment of the invention is comprised of a sequence of amino acids. The term amino acid as used in this invention is intended to include all L- and D- amino acids, naturally occurring and otherwise. Reagents comprising polybasic peptides provided by the invention include but are not limited to the following (the amino acids in the following peptides are L-amino acids except where otherwise indicated):

15      *acetyl-KC<sub>Acm</sub>GC<sub>Acm</sub>QAPLYKKIIKKLLES*  
*acetyl-KKC<sub>Acm</sub>GC<sub>Acm</sub>QAPLYKKIIKKLLES*  
*acetyl-KKKC<sub>Acm</sub>GC<sub>Acm</sub>QAPLYKKIIKKLLES*  
*acetyl-KKKKC<sub>Acm</sub>GC<sub>Acm</sub>QAPLYKKIIKKLLES*  
*acetyl-KKKKKC<sub>Acm</sub>GC<sub>Acm</sub>QAPLYKKIIKKLLES*  
*acetyl-KC<sub>Acm</sub>GC<sub>Acm</sub>GGPLYKKIIKKLLES*  
*acetyl-KKC<sub>Acm</sub>GC<sub>Acm</sub>GGPLYKKIIKKLLES*  
*acetyl-KKKC<sub>Acm</sub>GC<sub>Acm</sub>GGPLYKKIIKKLLES*  
*acetyl-KKKKC<sub>Acm</sub>GC<sub>Acm</sub>GGPLYKKIIKKLLES*  
*acetyl-KKKKKC<sub>Acm</sub>GC<sub>Acm</sub>GGPLYKKIIKKLLES*

20      Polybasic peptide embodiments of the present invention may be chemically synthesized *in vitro*. Peptides of the present invention can generally advantageously be prepared on a peptide synthesizer. The peptides of this invention can be synthesized wherein the radiolabel binding moiety is covalently linked to the peptide during chemical *in vitro* synthesis, using techniques well known to those with skill in the art. Such peptides covalently-linked to the radiolabel binding moiety upon synthesis are advantageous because specific sites of covalent linkage can be determined therein.

30      Radiolabel binding moieties of the invention may be introduced into the target specific

polybasic compound is a peptide and the peptide is synthesized by solid phase peptide synthesis.

18. The reagent of Claim 2 wherein the radiolabel binding moiety is covalently linked to the peptide during *in vitro* chemical synthesis.

19. The reagent of Claim 18 wherein the radiolabel binding moiety is covalently linked to the peptide during solid phase peptide synthesis.

5 20. A composition of matter having the formula:

*acetyl-KC<sub>Acm</sub>GC<sub>Acm</sub>QAPLYKKIICKLLES*

*acetyl-KKC<sub>Acm</sub>GC<sub>Acm</sub>QAPLYKKIICKLLES*

*acetyl-KKKC<sub>Acm</sub>GC<sub>Acm</sub>QAPLYKKIICKLLES*

10 *acetyl-KKKKC<sub>Acm</sub>GC<sub>Acm</sub>QAPLYKKIICKLLES*

*acetyl-KKKKKC<sub>Acm</sub>GC<sub>Acm</sub>QAPLYKKIICKLLES*

*acetyl-KC<sub>Acm</sub>GC<sub>Acm</sub>GGPLYKKIICKLLES*

*acetyl-KKC<sub>Acm</sub>GC<sub>Acm</sub>GGPLYKKIICKLLES*

*acetyl-KKKC<sub>Acm</sub>GC<sub>Acm</sub>GGPLYKKIICKLLES*

15 *acetyl-KKKKC<sub>Acm</sub>GC<sub>Acm</sub>GGPLYKKIICKLLES*

*acetyl-KKKKKC<sub>Acm</sub>GC<sub>Acm</sub>GGPLYKKIICKLLES.*

21. The composition of matter of Claim 20 that is labeled with technetium-99m.

22. A composition of matter comprising a composition comprising the reagent of Claim 20 and polysulfated glycan.

20 23. The composition of Claim 22 that is labeled with technetium-99m.

24. A method for imaging a site of inflammation within a mammalian body comprising mixing whole blood with an amount from about 1 microgram to 100 milligrams of a polysulfated glycan to form a mixture, and further adding to the mixture an amount from about 1 microgram to 100 milligrams of a composition of matter comprising a technetium-99m radiolabeled reagent according to Claim 1 to form a radiolabeled mixture, administering said radiolabeled mixture to an animal having a site of inflammation *in vivo*, and detecting a radioactive signal from the Tc-99m localized at the site of inflammation.

25. A composition of matter comprising a reagent according to Claim 24.